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THE QUALITY OF OIL EXTRACTED FROM PALM PRESSED FIBRE USING AQUEOUS ENZYMATIC TREATMENT

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ABSTRACT

A certain amount of crude palm oil (CPO) still remained in the palm pressed fibre (PPF) after going through the conventional milling processes. The use of aqueous enzymatic oil extraction (AEOE) was to recover the oil, hence, leading to the increase in oil extraction rate. With regards to the oil quality, the extracted PPF oil was analysed and compared to CPO using the MPOB Test Methods. From the results, the extracted oil contained high concentration of carotenoid (2750-2800 ppm) and vitamin E (930-950 ppm). Ironically, the quality was slightly lower than CPO, of which the values of deterioration of bleachability index (DOBI), free fatty acid (FFA) content and peroxide value (PV) were 1.12±0.12, 8.51±0.64% and 4.73±0.16 meq kg^{-1}, respectively. With the exception of linolenic acid (C18:3), the fatty acid composition (FAC) of extracted oil was similar to CPO. Despite the oil quality, the use of an enzymatic route has the potential for the extraction of phytonutrient-rich oil from PPF.

Keywords: palm pressed fibre, aqueous enzymatic oil extraction, oil quality.

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INTRODUCTION

Crude palm oil (CPO) is the oil extracted from the mesocarp of oil palm fruits normally by using a screw press. After pressing, the mesocarp fibres called palm pressed fibres (PPF) are used as boiler fuel in palm oil mills. Studies have shown that the PPF still contains about 4% to 5% residual oil, which has substantial amount of carotenoids, ranging from 4000 ppm to 6000 ppm; vitamin E, between 2400 ppm and 3500 ppm and sterols in the range of 4500 ppm to 8500 ppm (Choo *et al*., 1996).

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The solvent extraction is generally the most efficient method of oil extraction from PPF (Majid *et al*., 2012; Neo*h et al*., 2011; Ng *et al*., 2004; Choo *et al*., 1996; Goh *et al*., 1985). The residual oil from PPF could also be extracted by a combination of solvent and physical processes which include supercritical CO_2 (Harrison *et al*., 2006; 2008; Nik Norulaini *et al*., 2008), hexane cold extraction process (Neoh *et al*., 2011), low pressure solvent extraction and pressurised liquid extraction (Fiorella *et al*., 2015), hot compressed water extraction (Mohd Sharizan *et al*., 2016a) and sub-critical extraction (Mustapa *et al*., 2011; Che Yunus *et al*., 2015). However, compared to the enzymatic treatment (Noorshamsiana *et al*., 2013), the solvent-based methods of extraction are more drastic and unfriendly to the environment. In general, the use of enzyme to recover oil from PPF is benign to the environment (Sharma and Gupta, 2006; Shah *et al*., 2005; Mishra *et al*., 2005; Sharma *et al*., 2002; Rosenthal *et al*., 2001).

Of the total carotenes in the PPF oil, the value of α- and β-carotenes is about 90% (Choo *et al*., 1996). A study by Harrison *et al*. (2006) mentioned that the carotenes content in the oil extracted from fresh PPF ranged from 2900 to 3500 ppm using the supercritical-carbon dioxide (SC-CO_2) method. For the hexane soxhlet extraction method (Majid *et al*., 2012), the carotenes content obtained was 1877 ppm. From the sub-critical oil extraction method, the β-carotene obtained was 330 to 780 ppm (Mustapa *et al*., 2011) and 400 to 900 ppm (Che Yunus *et al*., 2015), respectively. Apart from the solvent-based extraction method, the use of hot water compressed extraction (HWCE) tended to produce β-carotenes in the range of 800 to 1000 ppm (Mohd Sharizan *et al*., 2016b).

Compared to other vegetable oils, palm oil has the highest content of tocotrienols (Ng *et al*., 2004; Gunstone *et al*., 1986; Slover, 1971). The concentration of vitamin E in the fresh PPF oil extracted by ethanol cold extraction was more than 4000 ppm (Ng *et al*., 2004), while, the hexane soxhlet extraction was 1153 ppm (Majid *et al*., 2012). The vitamin E extracted from the SC-CO_2 method was 1950 to 3500 ppm (Harrison *et al*., 2006). The vitamin E in the form of tocopherols and tocotrienols play an important role for health supplements (Ng *et al*., 2004). Moreover, the γ- and δ-tocotrienols are antioxidants as they prevent the oxidative deterioration and contribute to the stabilisation of oils and fats (Gapor, 1995; Piironen *et al*., 1986; Slover, 1971).

The value of deterioration of bleachability (DOBI) indicates the capability of bleaching and degumming of CPO during the refining process. For example, high DOBI value (3.24 and above) would result in cost-saving due to a reduction in phosphoric acid and bleaching earth consumption during refining process. Thereby, most of the natural antioxidants such as tocopherols and tocotrienols are retained in the refined oil (Siew, 1989). Based on Malaysian Standard (2007), the DOBI value of CPO should be 2.3 and above. Amongst the methods used, the SC-CO_2 technique seemed to give the DOBI value, ranging from 2.13 to 2.21 for PPF oil (Harrison *et al*., 2006).

The enzymatic hydrolysis of the oil is highly influenced by the presence of lipoids in oil palm fruits. In general, the free fatty acid (FFA) in CPO is formed due to autocatalytic hydrolysis of lipolytic enzyme inside the fruit. With regards to the quality of CPO, the value of FFA should be between 2.0% and 5.0%, whereas the FFA content of more than 5.0% will result in higher refining losses (Monday *et al*., 2000). A better quality of PPF oil was obtained from fresh PPF when subjected to SC-CO_2 extraction. The FFA value in the oil ranged between 3.46% and 3.88% (Harrison *et al*., 2006). In contrast, the PPF oil contained higher FFA would result in lower quality oil compared to CPO (Majid *et al*., 2012; Choo *et al*., 1996).

Apart from the FFA and DOBI values, the peroxide value (PV) (1.0 meq kg^{-1} peroxide and below) (Malaysian Standard, 2007) is also an important indicator to determine the degree of oxidation in CPO. The reaction of oxygen in the oil, which is catalysed by the presence of metals and high temperature used will cause an oxidative deterioration. Consequently, the reaction of unsaturated glycerides with oxygen will result in the formation of different chemicals such as aldehydes, ketones, hydro-peroxides and carboxylic acids. However, the PV analysis does not determine the secondary oxidation product such as aldehydes, hence, it does not indicate the actual oxidative deterioration in the oil (Siew, 2000). High quality PPF oil having PV ranging from 0.46 to 0.52 meq kg^{-1} was produced by SC-CO_2 extraction (Harrison *et al*., 2006). Nevertheless, Majid *et al*. (2012) reported that prolonged drying at high temperature of PPF prior to hexane sohxlet extraction resulted in PV of 8.5 meq kg^{-1}.

This article aims to determine the quality of extracted PPF oil using an aqueous enzymatic oil extraction (AEOE) process. Since the process employed mild conditions, it was anticipated that the quality of PPF oil is comparable to CPO.

MATERIALS AND METHODS

Materials

Fresh PPF was obtained from Palm Oil Mill Technology Centre (POMTEC) in Labu, Negeri Sembilan, Malaysia. The PPF was subjected to cellulase enzyme for the extraction of oil using the AEOE method, as described in previous study by Noorshamsiana *et al*. (2013).

Enzyme. The cellulase produced from *Trichoderma* reesei by Novo Nordisk, Denmark and commercially named Celluclast® 1.5L FG, has a declared activity of 700 EGU g^{-1} (EGU = Endo-Glucanase Units).

Oil extraction process. Enzymatic treatment was carried out using the following parameters; temperature of 55°C, pH of 5.6, 200 unit g^{-1} of enzyme, 100 ml of buffer solution, 100 rpm orbital shaker speed and 20 min of ultrasonification process prior 3 hr of enzymatic hydrolysis. Specific details explaining the whole process including the equipment and methodology used in the extraction process have already been published earlier (Noorshamsiana *et al*., 2013). The extracted oil was then stored at 5°C.

Analysis of the PPF Oil Quality

Carotene content. The carotene content was determined following the Method MPOB p2.6

(MPOB, 2005). Samples of PPF oil were diluted with an iso-octane (2, 2, 4-trimethylpentene). The mixture was analysed in a spectrophotometer at a wavelength of 446 nm (Hitachi, U-2800). The carotene content was calculated from the measurements of absorption.

Components of vitamin E. The vitamin E components in the oil were determined using high performance liquid chromatography (HPLC) (Hewlett Packard HP1100). The oil extract was diluted with hexane and a 20 μl sample was injected into the HPLC. For the analysis, the column used was silica column (150 mm ID x 6 mm length), mobile phase of 0.5% iso-propanol in hexane at a flow rate of 1 ml min^{-1} and identified using fluorescence detector.

DOBI. The DOBI was determined following the Method MPOB p2.9 (MPOB, 2005). Approximately, 100 mg of the oil sample was weighed and dissolved in 2, 2, 4-trimethylpentene iso-octane. The index was measured using the spectrophotometer (Hitachi, U-2800) with the absorbance at 446 and 269 nm.

FFA value. The FFA value was determined following the Method MPOB p2.5 (MPOB, 2005). The oil sample was melted at 60ºC to 70ºC and 50 ml of 0.1 N sodium hydroxide (NaOH) was added. The solution was then titrated with 0.5 ml of phenolphthalein at 40ºC until it reached the end-point (pink). The FFA value for the oil was calculated using Equation (1):

$$\text{FFA \%, as palmitic acid} = \frac{25.6 \times N \times V}{W} \quad \text{Equation (1)}$$

where N - normality of NaOH solution;
V - volume of NaOH solution in ml;
W - weight of the sample in g.

PV. The value of peroxide was determined following the Method MPOB p2.3 (MPOB, 2005). A total of 30 ml acetic acid with chloroform mixture was added into the oil sample. The solution was stirred and then added with 0.5 ml of saturated potassium iodide and 30 ml of distilled water. The mixture was titrated with 0.01 N sodium thiosulphate solutions with 0.5 ml starch added as a titration indicator, until it reached the end-point.

Fatty acid composition (FAC). For the FAC analyses, the triglycerides (TG) were converted to fatty acid methyl esters (FAME) first using sodium methoxide in petroleum spirit. The FAME was analysed using a gas chromatography (GC) (Perkin Elmer) following the Method MPOB p3.5 (MPOB, 2005). The prepared oil was injected into a GC with a Flame Ionisation Detector (GC-FID) system. A Perkin Elmer GC was fitted with a BPX-70 capillary column (25 m × 0.22 mm ID) and a flame ionisation detector. The temperature programme was set up from 120°C to 220°C with a constant increment at 12°C min^{-1}. Temperatures of both the injector and detector were maintained at 260°C. Helium was used as a carrier gas.

RESULTS AND DISCUSSION

The Oil Quality

Table 1 describes the quality of the enzymatic oil extract versus CPO. The DOBI value of the enzymatic oil extracts was 1.12 ± 0.12, whereas the DOBI value was 2.38 ± 0.23 for CPO. Based on the Malaysian Standard (2007), the DOBI value of more than 2.3 is classified as a good quality oil, thus the enzymatic oil extract is being categorised as oil of low quality (Baharin *et al*., 2001). This is probably due to the oxidation and hydrolysis caused by prolonged heating during the separation process at 60°C to 70°C hr. Besides, the oil quality is also affected by the duration of oil storage (<48 hr) prior to analysis (Chong, 1991). Adding to this, the oil extract also contained trace metal (such as iron and copper) from soil, fertilisers, as well as the contaminants from the processing equipment (Siew, 2000). These traces of metal tend to reduce the quality of oil and the degree of bleachability during refining (Chong and Gapor, 1983). Consequently, the presence of moisture in PPF has a tendency to promote the growth of microorganism (such as fungi and bacteria) and thus, reducing the quality of enzymatic oil extracts (Chong and Gapor, 1983).

With regards to the FFA value, the enzymatic extracted oil was 8.51 ± 0.64%, while CPO was only 3.47 ± 0.15%. Since the FFA value of CPO is lower than 5.0% (Malaysian Standard, 2007), the CPO is considered as good quality oil. During purification process, the use of high temperature tended to hydrolyse the triglycerides into partial glycerides (mono- and diglycerides), resulting in higher FFA value in the enzymatic oil extract. The degree of bleachability is highly dependent on the efficiency

TABLE 1. QUALITY OF ENZYMATIC OIL EXTRACT AND CRUDE PALM OIL

Samples	Physico-chemical content		
	DOBI value	FFA (%)	PV (meq kg^{-1})
Enzymatic oil extract	1.12 ± 0.12	8.51 ± 0.64	4.73 ± 0.16
Crude palm oil	2.38 ± 0.23	3.47 ± 0.15	0.27 ± 0.05

Note: Data reported are the means of three replicate analyses of independent samples.
DOBI – deterioration of bleachability index.
FFA – free fatty acid.
PV – peroxide value.

of water removal (Cowa*n et al*., 2012). The presence of metal-based impurities *(i.e*. iron and copper) will cause the oil to oxidise, hence, contributing to higher FFA value (Rohaya and Ma, 2001). The natural levels of iron and copper in CPO have been reported in the range of 0.5 – 1.0 ppm and 0.01 – 0.18 ppm, respectively (Chong and Gapor, 1983). Contamination during process and handling could have increased the amount of trace metals in oils. Moderate metal wear occurs during the pressing operation, creating a source of iron contamination. It was reported that the rate of iron contamination is 12.27 ppm t^{-1} of fresh fruit bunch processed (Bek-Nielson, 1969).

Compared to the CPO, the enzymatic oil extract had higher PV value (4.73±0.16 meq kg^{-1}). This is probably due to the reaction of metal-based impurities as pro-oxidants, which in turn, will promote the formation of peroxides (Siew, 1989). In order to produce good quality oil, the PV value should be below 2.0 meq kg^{-1} (Malaysian Standard, 2007).

Phytonutrients Content

The enzymatic oil extract had higher contents of β-carotene compared to CPO (*Table 2*). This indicates that some of the phytonutrients are still embedded inside the PPF cell-wall, even though after the extraction of CPO. The PPF cell-wall seemed to disintegrate when subjected to the enzymatic hydrolysis, hence releasing the phytonutrients (Hemavati and Jamaliah, 2015).

In contrast to previous findings that used solvent extraction to recover the PPF oil (Ng *et al*., 2004; Choo *et al*., 1996; Goh *et al*., 1985), the amount of vitamin E (α-tocopherol, α-tocotrienol, γ-tocotrienol and δ-tocotrienol) available in enzymatic oil extract was only in the range of 930 ppm to 950 ppm. This may be due to a lengthy process of 20 min of ultrasonication followed by 3 hr of enzymatic treatment, which in turn, will result in the oxidation and hydrolysis of the oil. Furthermore, the tocopherols and tocotrienols are highly prone to oxidation (Siti Khadijah *et al*., 2007).

As described in *Table 2*, the major constituent of vitamin E isomer in CPO and PPF oil extracts consisted of γ-tocotrienol (360-365 ppm) and α-tocopherol (460-465 ppm), respectively. These findings are consistent with the previous studies (Ng *et al*., 2004; Choo *et al*., 1996; Goh *et al*., 1985).

FAC

Table 3 gives the FAC of the CPO and enzymatic PPF oil extract. In general, the CPO had seven fatty acid components, while the enzymatic PPF oil extract had only six. The FAC, in descending order are as follows: the palmitic acid content in the CPO was 50.3% and enzymatic PPF oil extract was 49.5%; oleic acid in the CPO was 38.9% and enzymatic PPF oil extract was 39.5%; linoleic acid in the CPO was 6.0% and enzymatic PPF oil extract was 5.4% and followed by stearic acid. The amount of stearic acid in the CPO was 2.9% and that in enzymatic PPF oil extract was 3.1%. In general, the C18:1, C18:2 and C18:3 are known as monoglycerides, diglycerides and triglycerides, respectively. Interestingly, the enzymatic PPF oil extract did not contain linolenic acid as compared to CPO. This finding is in the agreement with study conducted by Neoh *et al*. (2011).

TABLE 2. PHYTONUTRIENTS CONTENT IN ENZYMATIC OIL EXTRACT AND CRUDE PALM OIL

Sample	Phytonutrients content (ppm)					
		Vitamin E				
	Carotene	α-tocopherol	α-tocotrienol	γ-tocotrienol	δ-tocotrienol	Total
Enzymatic oil extract	2 750-2 800	460-465	180-185	205-210	85-90	930-950
Crude palm oil	550-600	215-220	290-300	360-365	115-120	980-1 005

Note: Data reported are the minimum and maximum value of three replicate analyses of independent samples.

TABLE 3. FATTY ACID COMPOSITION OF CRUDE PALM OIL (CPO) AND ENZYMATIC PALM PRESSED FIBRE (PPF) OIL EXTRACT

Samples	Carbon number (%)						
	C_{12} Lauric acid	C_{14} Myristic acid	C_{16} Palmitic acid	C_{18} Stearic acid	$C_{18:1}$ Oleic acid	$C_{18:2}$ Linoleic acid	$C_{18:3}$ Linolenic acid
CPO	1.00	0.87	50.26	2.88	38.86	6.00	0.14
Enzymatic PPF oil extract	1.38	1.02	49.53	3.14	39.51	5.42	-

As shown in *Table 3*, the oil samples seemed to contain traces amount of lauric acid. It was noted that the component of lauric acid (C12) is generally present in crude palm kernel oil. This may be due to the use of high pressure setting that cause the nut to break. As a result, some crude palm kernel oil was released into the samples during pressing. Choo *et al*. (1996) reported that the oil extracted from PPF fibre was contaminated with about 30% of palm kernel. The amount of C12 component in CPO should be less than 0.5% (Malaysian Standard, 2007).

CONCLUSION

The aqueous enzymatic method used was able to produce the PPF oil extract with high concentration of phytonutrients. The composition of fatty acid in the PPF oil extract was similar to that in the CPO. However, values of DOBI and FFA do not meet the standard requirement due to the processing parameters used. One approach to improve the oil quality is through the size reduction of PPF and followed by the removal of impurities prior to enzyme treatment. For value recovery, researches should be focusing on the separation technique and the recycling of enzymes.

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REFERENCES

BAHARIN, B S; LATIP, R A; CHE MAN, Y B and ABDUL RAHMAN, R (2001). The effect of carotene extraction system on crude palm oil quality, carotene composition and carotene stability during storage. *J. Amer. Oil Chem. Soc. Vol. 78(8)*: 619-623. DOI: 10.1007/s11746-001-0354-4.

BEK-NIELSON, B (1969). Factors responsible for the development of peroxides during production and handling of palm oil. *Incorporated Society Planters Conference on Quality and Marketing of Oil Palm Product.* Kuala Lumpur. p. 86-105.

CHE YUNUS, M A; ZUHAILI, I; JEFFRY, J; MUHAMMAD SYAFIQ, H R; NOOR AZIAN, M and LIZA, M S (2015). Extraction of beta carotene from palm mesocarp via green sub-critical carbon dioxide. *Jurnal Teknologi Vol. 74(7)*: 35-39.

CHONG, C L (1991). An overview of the effect of milling process and storage on the quality of crude palm oil. *Proc. of the Seminar on Developments in Palm Oil Milling Technology and Environmental Management.* Genting Highlands, Pahang.

CHONG, C L and GAPOR, M T (1983). Effects of moisture and trace metals on oil quality. Workshop on Quality in the Palm Oil Industry. *PORIM Report*.

CHOO, Y M; YAP, S C; OOI, C K; MA, A N; GOH, S H and ONG, A S H (1996). Recovered oil from palm pressed fiber: a good source of natural carotenoids, vitamin E and sterols. *J. Amer. Oil Chem. Soc. Vol. 73 (5)*: 599-602. DOI: 10. 1007/ BF02518114.

COWAN, D; HOLM, D C and YEE, H S (2012). Reduction in free fatty acids in crude palm oil by enzymatic remediation. *J. Oil Palm Res. Vol. 24*: 1492-1496.

FIORELLA, P C T; SYLVIA, C A A; JANCLEI, P C; HELENA, T G; TÂNIA, F C and MEIRELES, M A A (2015). Pressurized liquid extraction and low-pressure solvent extraction of carotenoids from pressed palm fiber: experimental and economical evaluation. *Food and Bioproducts Processing Vol. 94(15)*: 90-100. DOI: http://dx.doi.org/10.1016/j.fbp.2015.01.006.

GAPOR, M T (1995). Palm vitamin E: a value-added tocotrienols-rich fraction (TRF) from palm oil. *Palm Oil Developments No. 22*: 7-17.

GOH, S H; CHOO, Y M and ONG, A S H (1985). Minor constituents of palm oil. *J. Amer. Oil Chem. Soc. Vol. 62(2)*: 237-240. DOI: 10. 1007/BF02541384.

GUNSTONE, F D; HARWOOD, J L and PADLEY, F B (1986). *The Lipid Handbook.* Chapman and Hall: London and New York.

HARRISON, L N L; CHOO, Y M; MA, A N and CHUAH, C H (2008). Selective extraction of palm carotene and vitamin E from fresh palm-pressed mesocarp fiber (*Elaeis guineensis*) using supercritical CO_2. *J. Food Eng. Vol. 84(2)*: 289-296. DOI: https://doi.org/10.1016/j.jfoodeng.2007.05.018.

HARRISON, L N L; CHOO, Y M; MA, A N and CHUAH, C H (2006). Quality of residual oil from palm pressed mesocarp fiber (*Elaeis guineensis*) using supercritical CO_2 with and without ethanol. *J. Amer. Oil Chem. Soc. Vol. 83(10)*: 893-898. DOI: 10. 1007/S11746-006-5043-9.

HEMAVATHI, S and JAMALIAH, M J (2015). Enhancement of palm oil extraction using cell wall degrading enzyme formulation. *Malaysian J. Analytical Sciences Vol. 19(1)*: 77-87.

MAJID, R A; MOHAMAD, A W and MAY, C Y (2012). Properties of residual palm pressed fibre oil. *J. Oil Palm Res. Vol. 24*: 1310-1317.

MALAYSIAN STANDARD (2007). MS 814:2007. *Palm Oil-Specification*. Second revision. ICS: 67.200.10. Department of Standards, Malaysia.

MISHRA, D; SHUKLA, A K; DIXIT, A K and SINGH, K (2005). Aqueous enzymatic extraction of oil from mandarin peels. *J. Oleo Science, 54(6)*: 355-359.

MOHD SHARIZAN, M S; NOOR AZIAN, M; YOSHIYUKI, Y; TOMOYA, T; MOHD AZIZI, C Y; MUSTAFA KAMAL, A A and HON, L L (2016a). Crude palm oil (CPO) extraction using hot compressed water (HCW). *Sep. Purif. Technol., 169*: 103-112. DOI: 10.1016/j.seppur.2016.06.001.

MOHD SHARIZAN, M S; NOOR AZIAN M; YOSHIYUKI Y; MUSTAPHA KAMAL, A A and CHE YUNUS, M A (2016b). Hot compressed water extraction curve for palm oil and beta carotene concentration. *IOP Conference Series: Earth and Environmental Science Vol. 36(1)*: 012045. DOI:10.1088/1755-1315/36/1/012045.

MONDAY, O A; SIMEON, C A and JONATHAN, M (2000). Quality attributes and storage stability of locally and mechanically extracted crude palm oils in selected communities in rivers and Beyelsea states Nigeria. *J. Plants Foods for Human Nutrition Vol. 55(2)*: 119-126. DOI: 10898481.

MUSTAPA, A N; MANAN, Z A; MOHD AZIZI, C Y; SETIANTO, W B and MOHD OMAR, A K (2011). Extraction of β-carotenes from palm oil mesocarp using sub-critical R134a. *Food Chem. Vol. 125(1)*: 262-267. DOI: https://doi.org/10.1016/j.foodchem.2010.08.042.

MPOB (2005). *MPOB Test Methods* (Aini, K; Siew, W L; Tan, Y A; Nor Aini, I; Tang, T S and Nuzul, A I eds.). MPOB, Bangi.

NEOH, B K; THANG, Y M; ZAIN, M Z M and JUNAIDI, A (2011). Palm pressed fibre oil: a new opportunity for premium hardstock? *International Food Research J., 18*: 769-773.

NG, M H; CHOO, Y M; CHUAH, C H and MOHD ALI, H (2004). Separation of vitamin E (tocopherol, tocotrienol and tocomonoenol) in palm oil. *J. Lipids Vol. 39(10)*: 1031-1035. DOI: 10.1007/s11745-004-1327-y.

NIK NORULAINI, N A; ANEES, A; FATEHAH, M O; ADEL ASHUR, S B; MOHD ZAIDUL, I S and MOHD OMAR, A K (2008). Sterilization and extraction of palm oil from screw pressed palm fruit fiber using supercritical carbon dioxide. *Sep. Purif. Technol. Vol. 60 (3)*: 272-277. DOI: 10.1016/j.seppur.2007.08.024.

NOORSHAMSIANA, A W; ASTIMAR, A A; NUR IZZATI, I; SALMIATON, A; NORHAFIZAH, A; SITI MAZLINA, M K and NOR FAIZAH, J (2013). Combined effect of operational variables on aqueous enzymatic oil extraction from palm oil pressed fibre. *J. Oil Palm Res. Vol. 25(2)*: 197-207.

PIIRONEN, V; SYVAOJA, E L; VARO, P; SALMINEN, K and KOIVISTOINEN, P (1986). Tocopherols and tocotrienols in cereal products from Finland. *Cereal Chem. Vol. 63(2)*: 78-81.

ROHAYA, M H and MA, A N (2001). Effect of temperature on the quality of fresh crude palm oil at different stages of processing. *Oil Palm Bulletin No. 43*: 31-37.

ROSENTHAL, A; PYLE, D L; NIRANJAN, K; GILMOUR, S and TRINCA, L (2001). Combined effect of operational variables and enzyme activity in aqueous enzymatic extraction of oil and protein from soybean. *Enzyme and Microbial Technology 28*: 499-509.

SHAH, S; SHARMA, A and GUPTA, M N (2005). Extraction of oil from *Jatropha curcas* L. seed kernel by combination of ultrasonication and aqueous enzymatic oil extraction. *Bioresource Technology, 96*: 121-123.

SHARMA, A and GUPTA, M N (2006). Ultrasonic pre-irrediation effect upon aqueous enzymatic oil extraction from almond and apricot seeds. *Ultrasonic Sanochemistry, 13*: 529-534.

SHARMA, A; KHARE, S K and GUPTA, M N (2002). Enzyme assisted aqueous extraction of peanut oil. *J. Amer. Chem. Soc., 79*: 215-218.

SIEW, W L (2000). Enhancement of oil quality. *Advances in Oil Palm Research*. Volume 2. MPOB, Bangi. p. 935-967.

SIEW, W L (1989). An overview of DOBI and the use of discriminant functions analysis for quality of crude palm oil. *PORIM Information Series No. 4.*

SITI KHADIJAH, A; NIK AZIZ, S; ABDUL GAPOR, M T and KAMSIAH, J (2007). Heating reduces vitamin E content in palm and soy oils. *Malaysian J. Biochemistry and Molecular Biology Vol. 15(2):* 76-79.

SLOVER, H T (1971). Tocopherols in foods and fats. *Lipids, 6:* 291-296.